The effect of integrin-linked-kinase regulation on chemokine secretions in thyroid cancer

Authors: Lyndsay Boyd, Samantha McCarty, PhD, Lawrence Shirley, MD
Institutions: The Ohio State Univeristy

MAGNET RECOGNIZED
AMERICAN NURSES CREDENTIALING CENTER

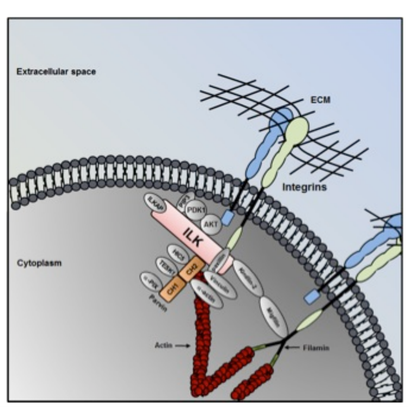
The James
The Ohio State University
Wexner Medical Center

Background

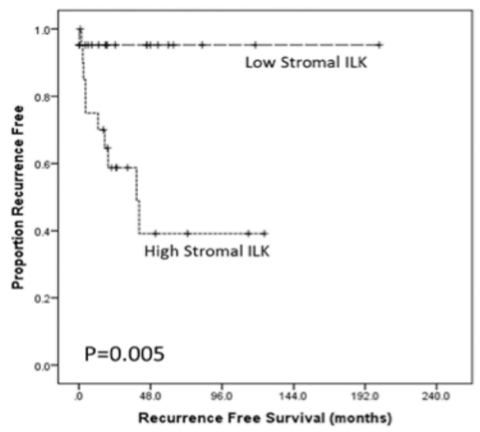
Over the past three decades, the incidence of thyroid cancer in the United States has been dramatically increasing. For patients with aggressive and undifferentiated forms, treatment options are limited and prognosis is poor. Predictors of death from thyroid cancer include the presence of gross local invasion and the development of distant metastases. It has been shown that the tumor microenvironment facilitates local invasion and metastasis through the recruitment of the immune system.

Although a variety of immune cells are recruited during carcinogenesis, including macrophages, natural killer cells, and lymphocytes, tumors are able to preferentially recruit sub-types that promote cancer growth, including tumor-associated macrophages and regulatory T cells. Small signaling molecules called chemokines, such as RANTES, IP-10, MCP-1, and MIP1-alpha, both attract pro-malignant immune cells as work to enhance the aggressiveness of cancer cells themselves. Our laboratory has previously shown a critical role of the protein integrin-linked kinase (ILK) in the aggressiveness of thyroid cancers, via promoting anchorage-independent growth, EMT, inhibition of apoptosis, and increased cell migration and invasion.

ILK Cellular Signaling



Increase in stromal ILK expression correlates with lymph node metastasis and shorter recurrence-free survival



Preliminary data showed that ILK was expressed in both the tumor tissue and surrounding stroma, however its expression in the stroma was more intense. Further evaluation of these samples showed that the increase in stromal ILK expression correlated with the presence of positive lymph nodes and also correlated with a shorter duration of recurrence-free survival (P=0.005).

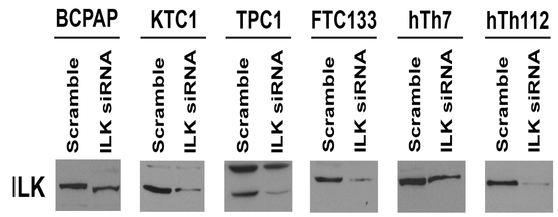
We thus investigated the role of ILK in secretion of chemokines from thyroid tumor as a possible mechanism for modulating the tumor stroma.

Material and Methods

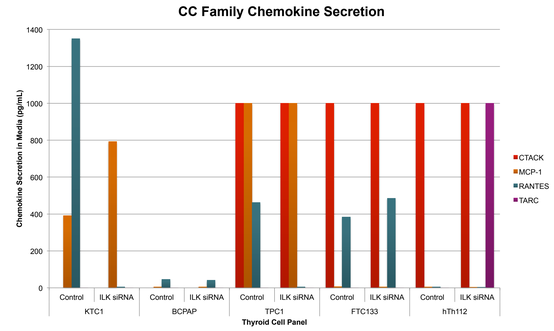
A panel of 5 human thyroid cancer cell lines (papillary cancer lines KTC1, TPC1, BCPAP; follicular cancer line FTC133; anaplastic cancer line hTh112) were transfected with either ILK or control siRNA, in triplicate. Cytokine 65-Plex Discovery Assays (Eve Technologies) were used to assess chemokine secretion. Western blots were completed on the 5 cell lines to evaluate ILK protein knock down in triplicate.

Results

Successful knock down of ILK expression in thyroid cancer cell line panel

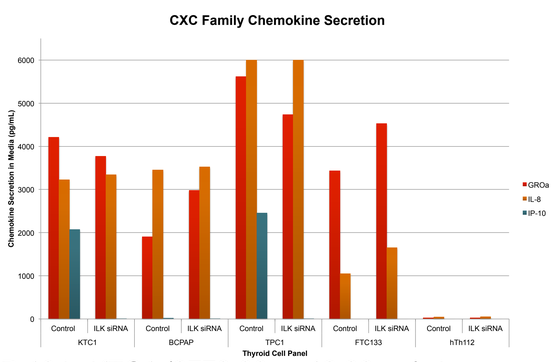


CC chemokine levels in ILK knockdown thyroid panel; decreased RANTES secretion, high secretion MCP-1 in PTC



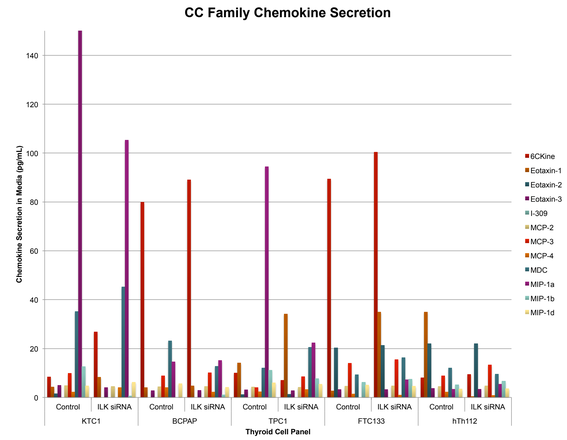
ILK inhibition led to significantly decreased secretion of RANTES in the cell lines KTC1 (1261 vs 6.8 pg/mL, P<0.01) and TPC1 (462.87 vs 5.58 pg/mL, P <0.01).

CXC Chemokine levels in ILK knockdown thyroid panel; decreased IP-10 in PTC



IP-10 in KTC1 (3773.06 vs 12.86 pg/mL, P<0.01) and TPC1 (2459.66 vs 2.71 pg/mL, P<0.01).

CC chemokine levels in ILK knockdown thyroid panel; decreased MIP-1a secretion in PTC



MIP1-alpha (KTC1, 223.69 vs 20.7 pg/mL, P<0.01; TPC1, 94.48 vs 22.33 pg/mL, P=0.01).

Conclusions

Successful knockdown of ILK protein expression in all five lines was confirmed via Western blot. ILK inhibition led to significantly decreased secretion of chemokines RANTES, IP-10, in KTC1, and MIP1-alpha in the papillary thyroid cancer cell lines KTC1 and TPC1. This established a new role for ILK in regulating the thyroid cancer microenvironment. Decreased secretion of chemokines in ILK knock down cell lines seems to be specific for papillary thyroid cancers. No significant changes were seen in anaplastic or follicular thyroid cancers.

Further research is needed to determine mechanisms by which ILK alters chemokine secretion and how this effects the types of immune cells that infiltrate the tumor microenvironment.

Acknowledgements

This project was funded by the Department of Education through the Undergraduate Research Summer Fellowship at The Ohio State University.